



GM2/GD2 Synthase siRNA (r): sc-270209

BACKGROUND

GM2/GD2 Synthase is a 533 amino acid protein encoded by the human gene B4GALNT1. The GM2 and GD2 gangliosides are sialic acid-containing glycosphingolipids that play a role in signal transduction and cell-cell recognition. GM2/GD2 Synthase is expressed abundantly in normal brain tissue of vertebrates. It contains a single 18 amino acid hydrophobic segment near the amino-terminus flanked by basic residues. GM2/GD2 Synthase primarily controls the balance between expression of simple and complex gangliosides at the cell surface. The ganglioside GD2 is expressed using GM2/GD2 Synthase in almost all neuroblastomas (NBs) as well as other neuroectoderm-derived tumor cells, such as malignant melanoma, adult T cell leukemia and some colon and gastric cancers. GM2/GD2 Synthase is a useful marker for NBs and may aid in evaluating adjuvant treatment efficacy in neuroblastoma with prognostic potential.

REFERENCES

1. Jacques, S., et al. 2005. Chemoenzymatic synthesis of GM3 and GM2 gangliosides containing a truncated ceramide functionalized for glycoconjugate synthesis and solid phase applications. *Org. Biomol. Chem.* 4: 142-154.
2. Marconi, S., et al. 2005. Expression of gangliosides on glial and neuronal cells in normal and pathological adult human brain. *J. Neuroimmunol.* 170: 115-121.
3. Saha, S., et al. 2005. Gangliosides enhance migration of mouse B16-melanoma cells through artificial basement membrane alone or in presence of Laminin or Fibronectin. *Indian J. Exp. Biol.* 43: 1130-1138.
4. Zhang, J., et al. 2005. Gangliosides activate the phosphatase activity of the erythrocyte plasma membrane Ca^{2+} -ATPase. *Arch. Biochem. Biophys.* 444: 1-6.
5. Wu, G., et al. 2005. Enhanced susceptibility to kainate-induced seizures, neuronal apoptosis, and death in mice lacking ganglioside GM1. *J. Neurosci.* 25: 11014-11022.
6. Dyatlovitskaya, E.V., et al. 2006. Role of biologically active sphingolipids in tumor growth. *Biochemistry* 71: 10-17.

CHROMOSOMAL LOCATION

Genetic locus: B4galnt1 (rat) mapping to 7q22.

PRODUCT

GM2/GD2 Synthase siRNA (r) is a pool of 3 target-specific 19-25 nt siRNAs designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10 μM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see GM2/GD2 Synthase shRNA Plasmid (r): sc-270209-SH and GM2/GD2 Synthase shRNA (r) Lentiviral Particles: sc-270209-V as alternate gene silencing products.

For independent verification of GM2/GD2 Synthase (r) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-270209A, sc-270209B and sc-270209C.

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20°C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20°C , avoid contact with RNases and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330 μl of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μl of RNase-free water makes a 10 μM solution in a 10 μM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

GM2/GD2 Synthase siRNA (r) is recommended for the inhibition of GM2 expression in rat cells.

SUPPORT REAGENTS

For optimal siRNA transfection efficiency, Santa Cruz Biotechnology's siRNA Transfection Reagent: sc-29528 (0.3 ml), siRNA Transfection Medium: sc-36868 (20 ml) and siRNA Dilution Buffer: sc-29527 (1.5 ml) are recommended. Control siRNAs or Fluorescein Conjugated Control siRNAs are available as 10 μM in 66 μl . Each contain a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA. Fluorescein Conjugated Control siRNAs include: sc-36869, sc-44239, sc-44240 and sc-44241. Control siRNAs include: sc-37007, sc-44230, sc-44231, sc-44232, sc-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

GENE EXPRESSION MONITORING

GM2/GD2 Synthase (C-5): sc-376505 is recommended as a control antibody for monitoring of GM2/GD2 Synthase gene expression knockdown by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500).

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgG κ BP-HRP: sc-516102 or m-IgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG κ BP-FITC: sc-516140 or m-IgG κ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor GM2 gene expression knockdown using RT-PCR Primer: GM2/GD2 Synthase (r)-PR: sc-270209-PR (20 μl). Annealing temperature for the primers should be $55-60^{\circ}\text{C}$ and the extension temperature should be $68-72^{\circ}\text{C}$.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.